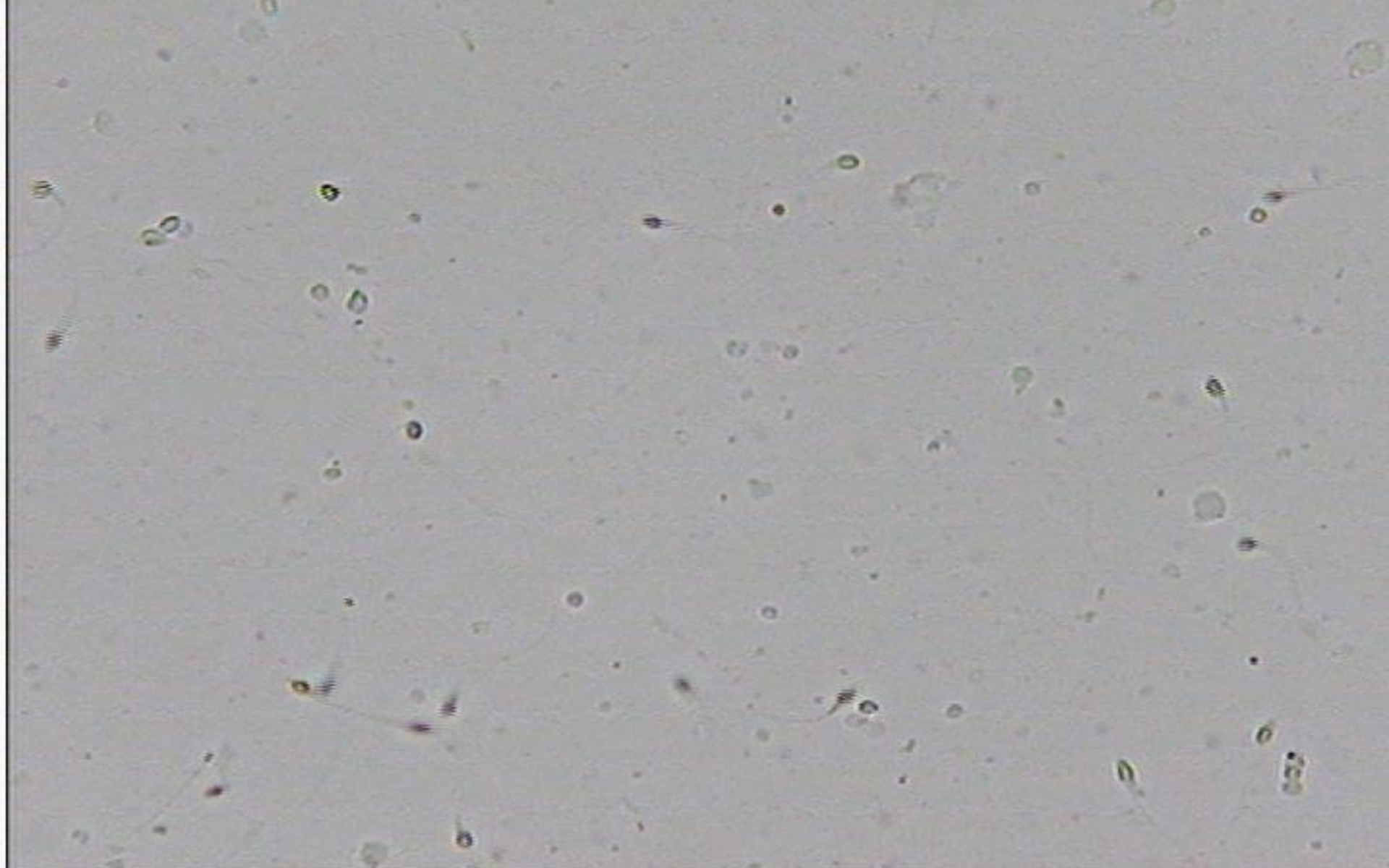
Semen parameters lab. & clinical interpretation

BY
Dr. Attia Abdalla Attia
Prof. of
Dermatology & Andrology
Al- Azhar Faculty of medicine – Cairo.




(sperm from fertile semen)

Semen

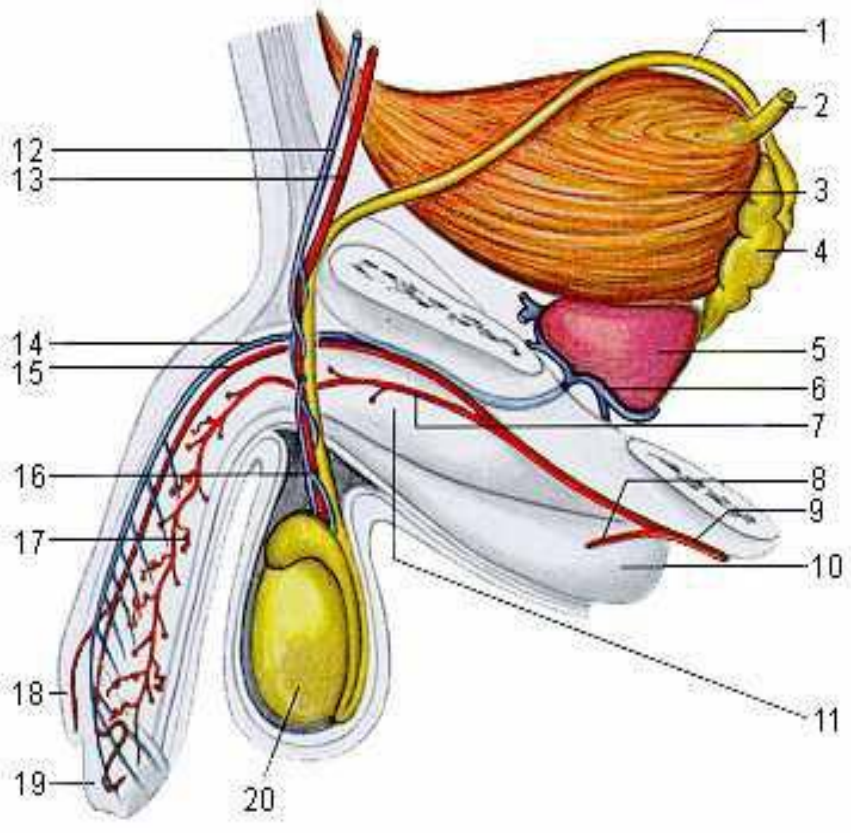
The Semen consists of two components:

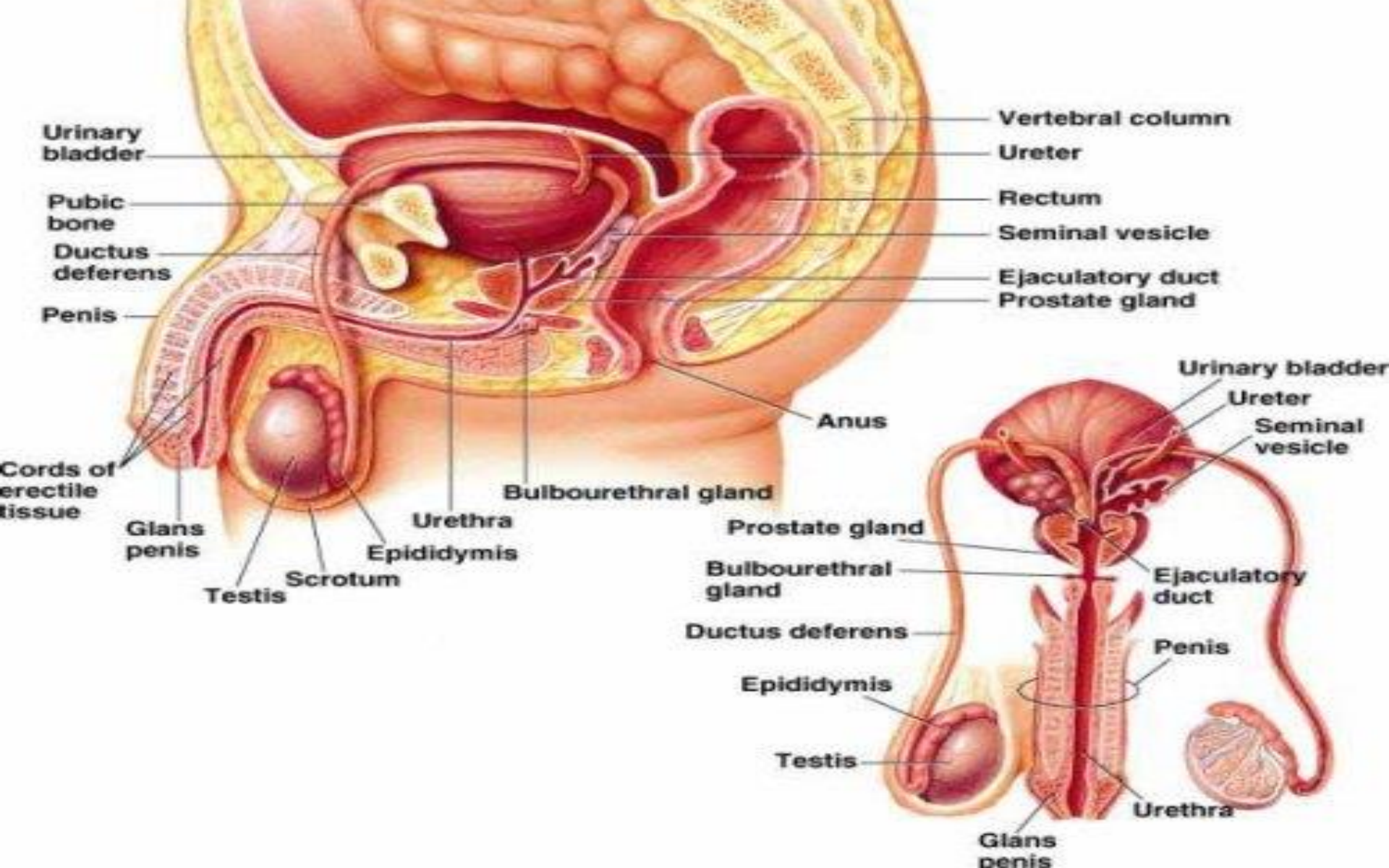
1) Seminal plasma

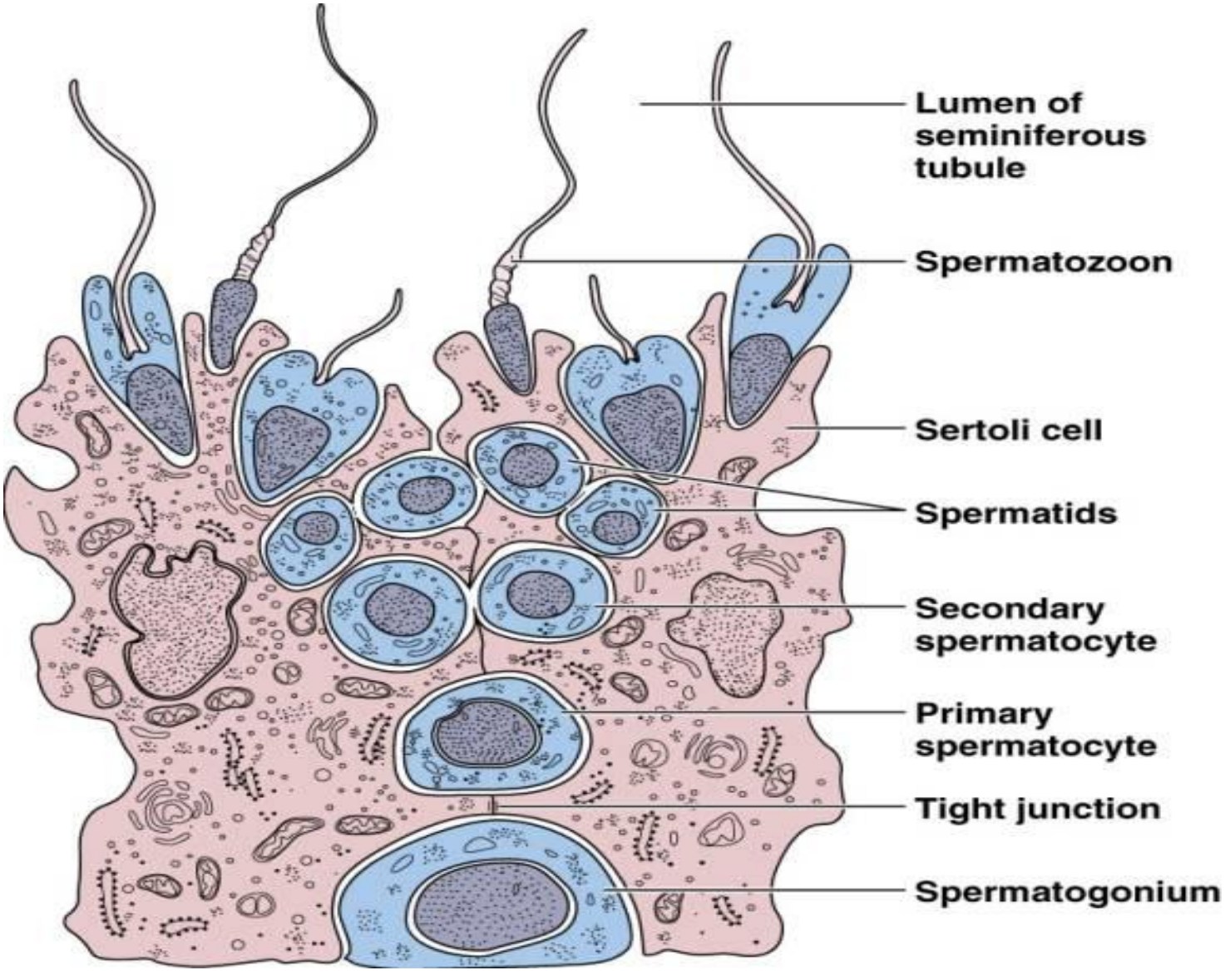
-SV. (65%) -prostate (30%)

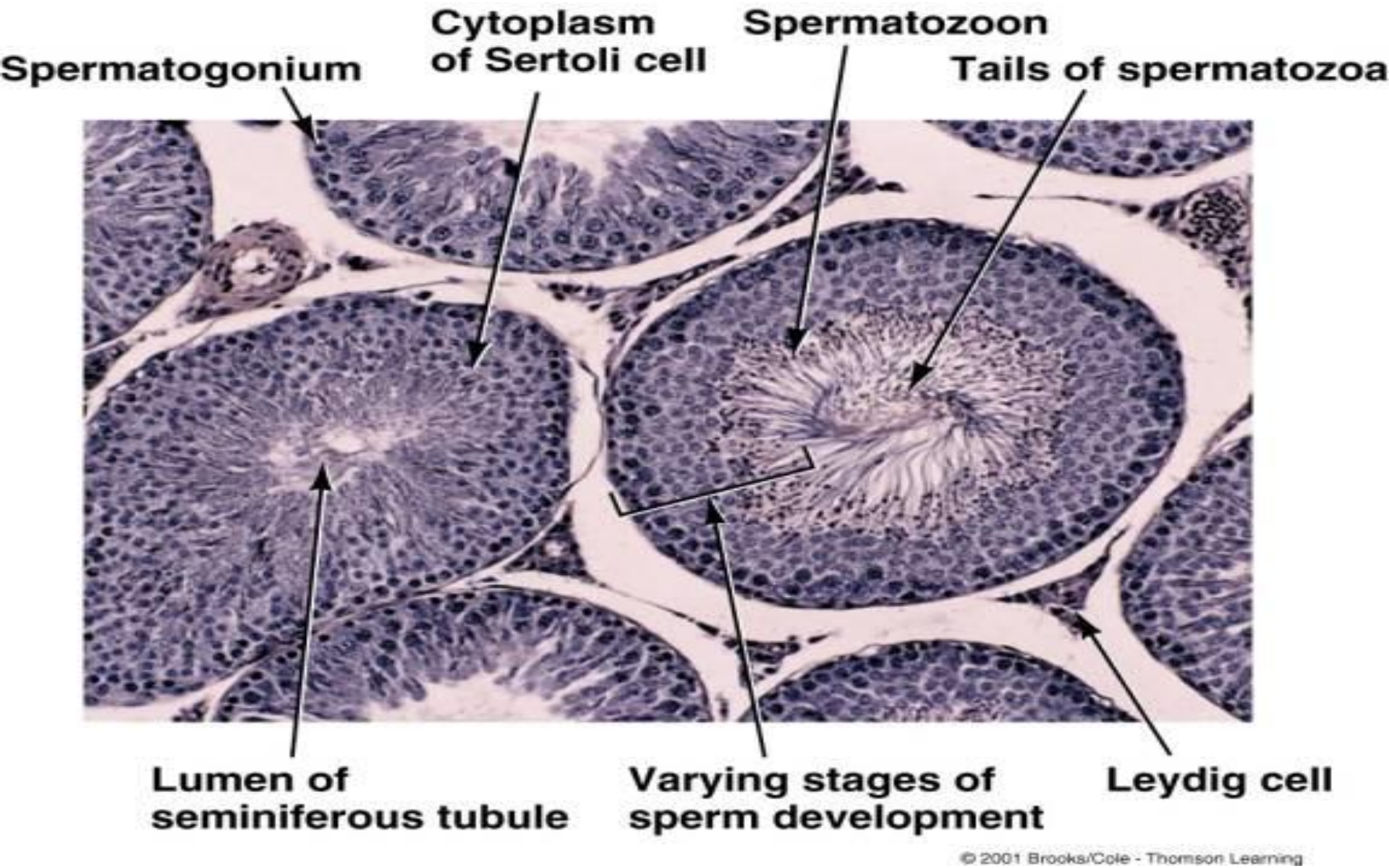
-the other accessory glands(5%)

2) Sperms in the seminiferous tubules




Urinary bladder
Pubic bone
Ductus deferens
Penis
Cords of erectile tissue
Glans penis
Testis
Scrotum
Epididymis
Urethra
Bulbourethral gland
Anus
Vertebral column
Ureter
Rectum
Seminal vesicle
Ejaculatory duct
Prostate gland
Urinary bladder
Ureter
Seminal vesicle
Prostate gland
Bulbourethral gland
Ejaculatory duct
Ductus deferens
Penis
Epididymis
Testis
Urethra
Glans penis


Lumen of seminiferous tubule
Spermatozoon
Sertoli cell
Spermatids
Secondary spermatocyte
Primary spermatocyte
Tight junction
Spermatogonium



© 2001 Brooks/Cole - Thomson Learning

Semen analysis

- **Semen analysis must include these points :**

1-Name

2-Date

3-Duration of abstinence

Semen analysis

Macroscopic characters :

1-Volume of semen sample

2-Odor,

3- Color,

4- PH,

5-Liquefaction time

6-Viscosity

Semen analysis *(cont.)*

Microscopic characters:

1 -Count of sperms /ml.

2 -Count of sperms/ejaculate

3 -Viability of sperms

4 -Motility of sperms

5-Abnormal forms

6-Aglutination

7-RBC's

8-Round cells(germinal cells & pus cells)

When can you judge on the semen parameters?

- One semen analysis is not enough
- 2 analysis in cases of oligozoospermia
- 3 analysis in cases of azoospermia
- At least 2 weeks between each one
- Recent analysis should be available

Length of abstinence:

- A normal male produces 80 mil.sperm /day.
- A short *abstinence* reduces sperm number Prolonged *abstinence* reduces the motility.
- Some feel that the *abstinence* period must be according to the patient's normal coital frequency.
- WHO recommend 2-7 days .

Transport of a semen sample to the laboratory:

@ < ½ an hours

@The sample should be protected from extremes of:

-temperature < 20^0C & > 40^0C

-direct sun rays

-vigorous movement

THE SEMEN VOLUM

- **An adequate volume is required to:**

- **transport sperm into the female reproductive tract**
- **allow for fertilization of the oocyte.**

Matthew et al, (December 2009)

CUAJ (3), 6

OLIGOSPERMIA

LOW VOLUM <1.5ML

THE CAUSES ARE MANY(ej.d.,.. +

RETROGRADE EJACULATION

IS there specific number of sperms should be found in urine to said , it is +ve retrograde?

POST COITAL URINE

- The prevalence of sperm in the post-ejaculatory urine in the fertile population is similar to that in infertile patients,

Urology. 2008 Jan;71(1):110-2.

Therefore the presence of sperm in urine does not mean retrograde ejaculation except in cases of azoospermia

CUAJ ,December 2009

WHEN WE CAN SAY IT IS +ve RETROGRADE EJACULATION ?

-Total Sperm Number in Urine is **>** 3.8×10^6 and the Retrograde Index (RI) IS **>** 2.16%.

Arch Androl.2005 Nov-Dec;51(6):431-6.

-The presence of sperms in cases of AZOOSPERMIA

CUAJ ,December 2009

CAUSES OF RETROGRAD EJACULATION

- 1- Diabetes,
- 2- Incomplete bladder neck cooptation
- 3- POST

  -transurethral surgery,

  -retroperitoneal lymph nodes dissection,

  -spinal cord injuries.

The color of seminal fluid

- A normal semen has a homogenous, **grayish white** (high protein content)
- **urine in semen**

  a **faint yellow** color, & detected by the uriniferous odor
- **Yellow color** in :Jaundice ,carotinemia ,drug
- **Blood in semen : hematospermia.**

Blood in semen : heamospermia.

-Caused by

(infection bacterial or parasitic eg.
bilharzial seminal vesiculitis,
ductal obstruction, bl. abnormalities &malignancy)

-Color depend on the amount and the age of blood

-Small amounts----------pink –

- Large quantities----------bright red –

-If the blood has been for some time----brown. –

The PH of semen:

@The PH must be measured within one hour of ejaculation

@The pH is determined by:

acidic secretions of the **prostate**

alkaline secretions of the **seminal vesicles**

- **Normal pH** 7.2 - 8.0

Abnormalities of The PH

Low pH + normal vol.

INFECTION(PYOSPERMIA)

Low PH +low vol.+ non coagulation

- CONG. ABSCENT VAS(SV)
- ED OBSTRUCTION
- PARTIAL RETROGRADE EJACULATION

HIGH PH (alkaline)

Chronic infection of the prostate(decreased its acidic products)

Coagulation

- At ejaculation,semen is a **liquid** ,

  BUT after contacting the SV secretion (the latter fraction of ejaculation), it **coagulates** immediately .

- The fibrinogen-like protein eg. **semenogelin** (**SV**)

  under the effect of **vesiculase** enzyme (**the prostate**) coagulation occur

Coagulation

- **Coagulation** allow:

  all sperms to be in contact with the ingredients of coagulum .

- **Failure of Coagulation**

  associated with:

  - decreased activity & or absent SV.gland.
  - poor sperm motility.

Liquefaction

- Liquefaction occurs within:
  5 minutes in vivo &
  20–30 minutes in vitro
- **Liquefaction** by :
  prostatic **proteolytic** enzymes
  (chymotrypsin-like, PSA, semenin pepsinogen ,amylase & hyaluronidase).
- Both **Coagulation** and **liquefaction** are induced by prostatic enzymes
- **DELAYED LIQUEFACTION :**
  Poor prostatic secretions (infection)

PROSTATE & SV

- **Coagulation ---- by**

the effect of **prostatic vesiculase** on

the fibrinogen-like proteins (**SV**)

(semenogelin)

- **liquefaction** ----- **by**

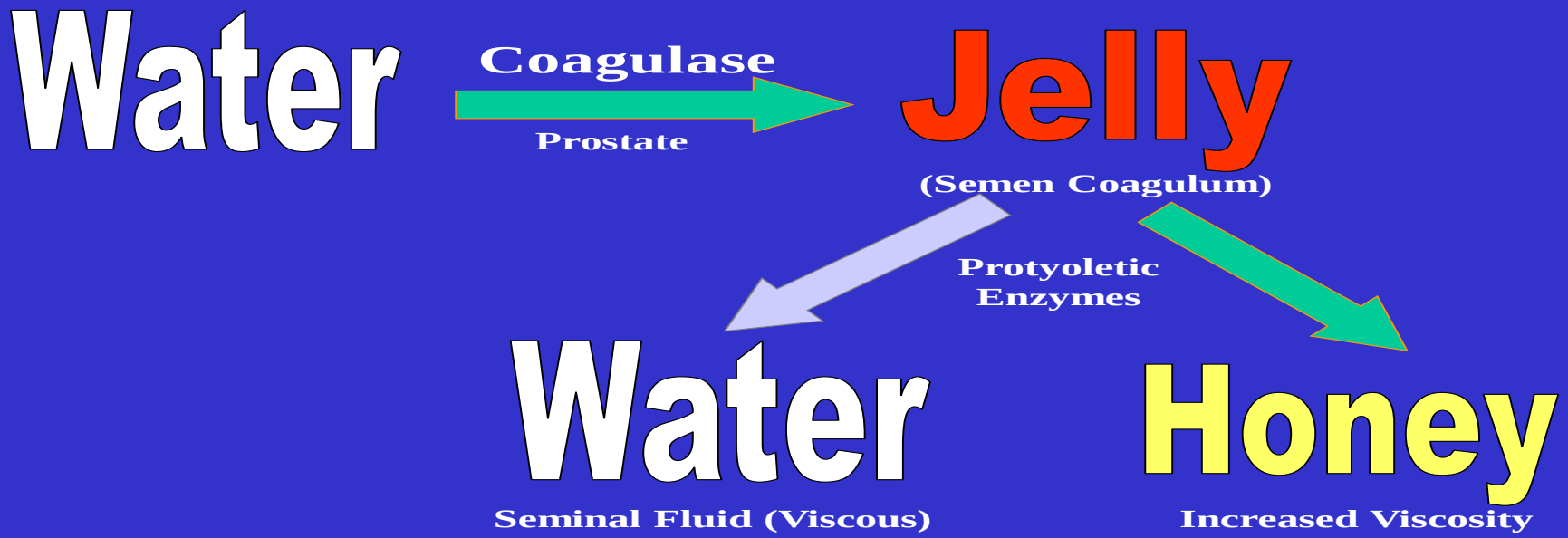
prostatic **proteolytic** enzymes eg.

(chymotrypsin-like, PSA, semenin)

SEMEN(FLUID)--------coagulation coagulum (JELLY)

liquefaction ----→ H.viscous (HONY)

→ fluid



The viscosity of semen

- The consistency = viscosity & not **coagulation**
- **The viscosity measured after liquefaction**
- **High viscosity**

associated with:

- **SV hypofunction**
- decreased sperm motility
- infertility (sperm unable to exit from seminal plasma to enter the cervical mucous).

viability

Viability must be >58% by dye

Vital stain(Eosin-Nigrosin stain)

to differentiate viable immotile sperms from immotile dead .

The hypo-osmotic swelling (HOS) test

- for viability &measures sperm membrane integrity which is relevant to fertilizing ability
- to select viable sperm for ICSI especially when there are no motile sperm in TESE or cryo. samples

Motility&viability

- If motility <5%-10%

viability testing is recommended

(low motility may be due to dead sperms)

- **Low motility% with a high % of viable sperms**

may reflect :

structural & metabolic abnormalities of sperms

due to:

@abnormalities in testicular function or

@antimotility factors in the seminal plasma

Motility Grads

(a) Progressive motility >32%

(rapid and slow or sluggish motility)

(b) **Progressive and non-progressive motility. (>40%)**

This will be within 60 min. of ejaculation

MOTILITY

Sperm motility depends on :

- **Normal spermatogenesis**
- **Physiologic epididymal sperm maturation**
- **Prompt ductile transport of spermatozoa**
- **Normal interrelationship of sperms with S.V. & prostatic secretion**

Sperm motility enhancers

Examples of motility enhancer in the semen

Stimulators products of the **SV** :

potassium, bicarbonate, magnesium,

PGE ,PGF , and prolactin

HOW

Bicarbonate stimulates sperm motility?

Activates adenylate cyclase leading

to increasing of cAMP

Sperm motility inhibitors (SPMI)

Examles of (SPMI) in the semen are :

@From **(SV)** the SPMI is found in the precursor form of **semenogelin** ,

BUT if degraded into smaller peptides by prostatic proteases shortly after ejaculation this SPMI effect disappeared.

@ Other (SPMI) :

ROS , pus cells , ASA , mycoplasma ,E coli , foreign bodies eg. water & soap

Causes of Low Sperm Motility

- Varicocele
- Hormonal imbalance,
- chronic infection .
- Nutrition & Vitamin Deficiency : vitamin C, selenium, zinc, and folate
- Chemotherapy,

Causes of Low Sperm Motility

- **Obesity,**
- **Smoking, Alcohol drinking,**
- **Exposure to Heavy metals**
- **Genetic Factors like Cystic fibrosis, Kartagener syndrome .**
- **Mental stress**

Non sperm cells

- **RBCs**
- **Epithelial cells**:-uretheral
  -vaginal in coitus interruptus
- **Round cells** :-Spermatogenic (germinal) cells
  - white blood cells (WBC).

To differentiate round cells:

1-Peroxidase test.

2- Leukocytes specific antigen

- BY Peroxidase test.

  -Leukocytes are brown color,

  -Other cells(epithelial & germinal) are pink.

NB : Leukocytes (mainly neutrophils)

Increased **spermatogenic** cells suggests :

- inadequate spermatogenesis

-premature release of:

spermatids,

spermatocytes or

spermatogonia

Pyospermia
Leukocytospermia

Pus cells: > 1 mil/ml

> 5/hpf

due to inflam. of the accessory glands

The Leukocytes produce 3 ROS :

superoxide anion, **hydrogen peroxide**,

and the **hydroxyl radical**

THESE (ROS) inhibit sperm motility and function.

NB:INFLAMMATION OF ACCESSORY GLANDS

may not associated with leukocytospermia

Agglutination *VS.* adherence

- **Agglutination** means **motile spermatozoa** only
  stick to each other,
  it is site-specific

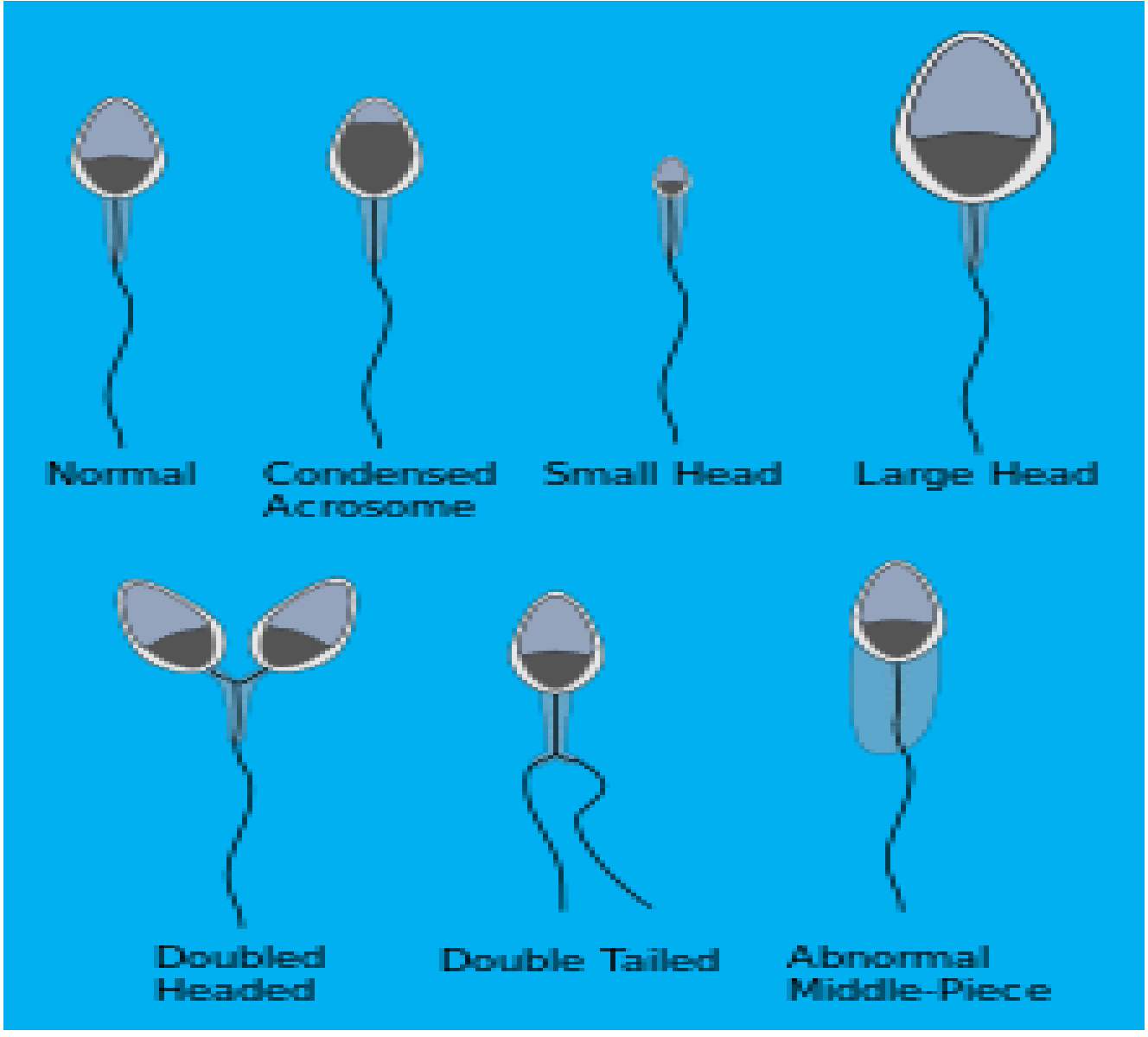
Agglutination = immunological fertility (ASA)

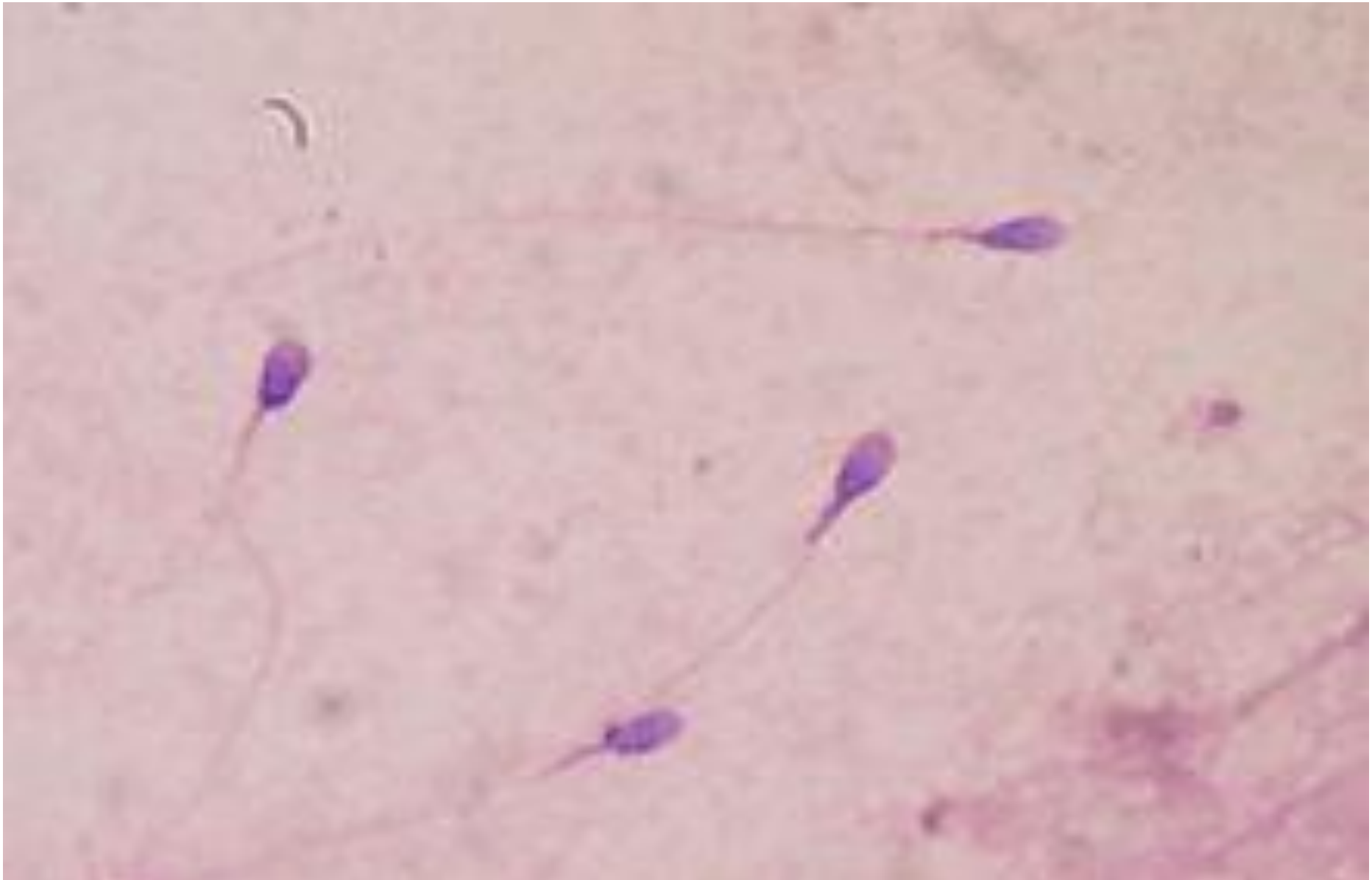
- ***BUT*** The **adherence** of **motile** or **immotile** spermatozoa to:
- -mucus threads
  - other cells
  -debris

it is non-specific agglutination indicates infection

Sperm Morphology

- The normal sperm morphology was 50% according to the **shape .**
- **BUT NOW** , the Sperm Morphology depends on the **morphometric** measurements (length, width, and length-to-width ratio) and the area occupied by the acrosome
- Therefore, it became highly predictive for the results of IVF.


Normal
Condensed Acrosome
Small Head
Large Head
Doubled Headed
Double Tailed
Abnormal Middle-Piece



Teratozoospermia

- **Abnormal forms**

  <70% WHO 3rd. Ed.,(1992)

  <86% WHO 4th. Ed.,(1999)

  (**Strict KRUGER Criteria)**

  <96% (WHO 2010)

Causes of teratospermia

- unknown in most cases.
- Defective spermatogenesis
- Varicocele
- Hodgkin's disease,
- coeliac disease
- Crohn's
- Infection

Effects of varicocele on :

1) Semen analysis :

- ↓ motility
- ↓ sperm count 20 mil. (65 %)
- ↑ abnormal forms

N.B

Stress pattern is not specific for varicocele.

- **CAN VARICOCELE LEAD TO AZOOSPERMIA?**

Varicocelectomy for

Functional Azoo with varicocele

can lead to return of sperms to

the ejaculate in 21 – 47 %.of cases

(Matthev's et al., 1998)

Effects of varicocele

- CAN AFFECT ONE PARAMETER ONLY ?
- IF YES
- CAN THIS EFFECT CONTINUE ,WITH TIME, WITHOUT AFFECTION OF THE OTHER PARAMETERS ?

Varicocelectomy

Post Operative Improvement:

- Sperm Motility 70%
- Sperm Density 51%
- Sperm Morphology 44%
- Conception Rate 40-50%

(Sigiman & Jarow ,2oo2)

- **The operation spare 50 % of azoospermic men from TESE (due to sperm production)**

- **Bilateral varicocelectomy before IVF improve success rate.**

(Gat et al., 2005)

Mobile phones and semen

- A mobile phone in talk mode in the pocket has:
  a risk if used for long-term.

Los Angeles Times study (September , 2008)

- In an *in vitro* pilot study by Agarwal *et al(2009)*

Exposure of sperms to cellular phone waves lead to:

↓ sperm motility and viability, ↑ ROS level.

Fertility and Sterility (2009)92 (4): 1318–25

SMOKING

- **Tobacco** smoking lowers the sperm quality, Smoking **marijuana** has similar effects

Wegner ,et al. (September 2009)

Fertil. Steril..

Macroscopic examination

	WHO 1999	WHO 2010
Volume	**2 ml or more**	**1.5 ml**
Odour	**Semeniferous**	
Colour	**Grayish**	
PH	**7.2 or more**	
Liquefaction	**Within 60 minutes**	
Viscousity	**Thread < 4 cm**	**Thread < 2 cm**

Microscopic examination

	WHO 1999	WHO 2010
Count	20 millionn/ml or more	15 million/ml or more
Total count	40 million or more	39 million or more
Viability	>75 %	>58 %
Motility	50% or > (a+b) or 25% or > (a)	32% or > (a+b) 40% or > (a+b+c)
abnormal forms	< 86%	< 96%
Pus cells	< 1 million/ml	< 1 million/ml

SEMEN Abnormalities(who,2010)

volume	< 1.5 ml	oligospermia
	> 5 ml	ploysperm ia
	No ejaculate	Aspermia
Count	<15 mil / ml	Oligozoospermia
	>250 mil / ml	Polyzoospermia
	No sperms	Azoospermia

SEMEN Abnormalities

Motility	**progressive < 32%** **Or progresive and non progressive <40%**	**Asthenozoospermia**
Abnormal forms	**96% or more**	**Teratozoospermia**
All sperms dead		**Necrozoospermia**
Pus cells	**> 1 mil/ml**	**Pyospermia**

Computer assisted sperm analysis CASA

- It is a semiautomated technique
- As regard sperm count ,the sample preparation & frame rate can affect accuracy of the CASA
- Stains used also have affected the accuracy of determining morphology
- **Although** this technology had theoretic advantages **but** these are not true in clinical practice

Semen Analysis

- **Semen analysis assess only production of the sperms and not their functions.**
- It was shown that 30% of all patients with normal semen analysis have abnormal sperm function.

- **Therefore , a normal semen analysis does not necessarily reflect normal fertility potential .**

THANK

YOU